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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/549,871	09/16/2005	Andreas Kunkel	7003/55	2451
27774 7590 08/06/2008 MAYER & WILLIAMS PC 251 NORTH AVENUE WEST 2ND FLOOR WESTFIELD, NJ 07090				
EXAMINER MEAH, MOHAMMAD Y				
ART UNIT		PAPER NUMBER		
1652				
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**Please find below and/or attached an Office communication concerning this application or proceeding.**

The time period for reply, if any, is set in the attached communication.

### Office Action Summary

**Application No.**

10/549,871

**Applicant(s)**

KUNKEL ET AL.

**Examiner**

MD. YOUNUS MEAH

**Art Unit**

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**Period for Reply** -- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

**Status**

- 1) ☒ Responsive to communication(s) filed on 24 May 2008.
- 2a) ☒ This action is **FINAL**. 2b) ☐ This action is non-final.
- 3) ☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

**Disposition of Claims**

- 4) ☒ Claim(s) 1-30 is/are pending in the application.
- 4a) Of the above claim(s) 11-14, 19-22, 25-28 and 30 is/are withdrawn from consideration.
- 5) ☐ Claim(s) \_\_\_\_\_ is/are allowed.
- 6) ☒ Claim(s) 1-10, 15-18, 23, 24 and 29 is/are rejected.
- 7) ☐ Claim(s) \_\_\_\_\_ is/are objected to.
- 8) ☐ Claim(s) \_\_\_\_\_ are subject to restriction and/or election requirement.

**Application Papers**

- 9) ☐ The specification is objected to by the Examiner.
- 10) ☐ The drawing(s) filed on \_\_\_\_\_ is/are: a) ☐ accepted or b) ☐ objected to by the Examiner.  
Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).  
Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).
- 11) ☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

**Priority under 35 U.S.C. § 119**

- 12) ☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).
- a) ☐ All b) ☐ Some \* c) ☐ None of:
1. ☐ Certified copies of the priority documents have been received.
  2. ☐ Certified copies of the priority documents have been received in Application No. \_\_\_\_\_.
  3. ☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

\* See the attached detailed Office action for a list of the certified copies not received.

**Attachment(s)**

- 1) ☐ Notice of References Cited (PTO-892)
- 2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
- 3) ☐ Information Disclosure Statement(s) (PTO/SB/888)  
Paper No(s)/Mail Date \_\_\_\_\_
- 4) ☐ Interview Summary (PTO-413)  
Paper No(s)/Mail Date \_\_\_\_\_
- 5) ☐ Notice of Informal Patent Application
- 6) ☐ Other: \_\_\_\_\_

### **DETAILED ACTION**

A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after allowance or after an Office action under *Ex Parte Quayle*, 25 USPQ 74, 453 O.G. 213 (Comm'r Pat. 1935). Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, prosecution in this application has been reopened pursuant to 37 CFR 1.114. Applicant's submission filed on 5/20/08 has been entered.

Claims 1-30 are pending. Claims 1-10, 15-18, 23-24 and 29 were examined in the previous action. Claims 11-14, 19-22, 25- 28 and 30 remain withdrawn. With supplemental amendment of this application, the applicant, on dates on 5/20/08, amended claims 1, 24, 29 and cancelled claims 31-32.

### ***Claim Rejections***

Applicants' arguments filed on 5/20/08, have been fully considered and are not deemed to be persuasive to overcome some of the rejections previously applied. Rejections not reiterated from previous office actions are hereby withdrawn.

**35 U.S.C 112 1ST paragraph Rejections**

*Written Description requirement Rejections:*

The following is a quotation of the first paragraph of 35 U.S.C. 112:

The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-10, 15-18, 23-24, 29 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

Claims 1-10, 15-18, 23-24, 29 are directed to a process of producing ergosta-5,7 dienol by culturing organisms having a decreased  $\Delta 22$ -desaturase activity wherein the  $\Delta 22$ -desaturase gene is inactivated by any type of mutagenesis, and an increased HMG-CoA reductase activity and increased activity of the squalene epoxidase of SEQ ID NO:8. or a variant thereof from any source. The specification fails to describe in any fashion the physical and/or chemical properties of the claimed class of ergosterol-biosynthetic enzymes including squalene epoxidase other than its involvement in ergosterol biosynthetic pathway. No relation between the structure of the species and function is described. Neither the type of organisms nor the structure of genes expressed in the organism are defined in the specification. The specification discloses only a few DNA of the claimed genus of  $\Delta 22$ -desaturase, HMG-CoA reductase and squalene epoxidase (i.e., SEQ ID Nos : 2, 4, 8 ) expressed in the

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microorganism for the production of few ergosterol compounds ( i.e., lanosterol, Zymosterol, 4,4 dimethyl-zymosterol), which are insufficient to put one of skill in the art in possession of the attributes and features of all species within the claimed genus. Therefore, one skilled in the art cannot reasonably conclude that the applicant had possession of the claimed invention at the time the instant application was filed.

Applicants argue that the claims require for their methods of production of ergosta-5,7 dienol, a organisms having increased expression of squalene epoxidase and HMG-CoA reductase and reduced activity of  $\Delta 22$ -desaturase and these genes in organisms can be modulated by standard genetic methods. They further argue that specification discloses in page 5-6 several of these methods (introducing nucleic acid sequences that inhibit desaturase activity, mutation, binding factors to inhibit, etc). However, this is not persuasive because the scope of variant of squalene epoxidase of SEQ ID NO: 8, the HMG-CoA reductase and of  $\Delta 22$ -desaturase gene to be modified encompassed is undefined.

Furthermore, there are many ways expression of squalene epoxidase and HMG-CoA reductase can be increased and  $\Delta 22$ -desaturase can be decreased in an organism. As the structures of all of the squalene epoxidases, HMG-CoA reductases and  $\Delta 22$ -desaturases that recited in the instant claims are not defined in any way, one of ordinary skill in the art would not be able to make and use any microorganism comprising said enzymes and would require undue experimentation to find out which of these mutant microorganisms comprising increased expression of squalene epoxidase and HMG-CoA reductase and

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decreased expression of desaturase will produce said compound. The specification fails to describe how a genus of microorganism can be transformed by any means to increase expression squalene epoxidase and HMG-CoA reductase and decrease expression of any  $\Delta^{22}$ -desaturase. Moreover, there are many ways in which a microorganisms' desaturase genes can be inactivated such as deletion, substitution of specific amino acid residues of DNA sequence or deletion of the whole genes, etc addition of inhibitors, modification of endogenous modulators, etc which would require substantial additional information that has not been provided by the specification. The disclosed species of squalene epoxidase and HMG-CoA reductase (SEQ ID NO:8 and SEQ ID NO:4) are not representative of any variant of SEQ ID NO:8 and SEQ ID NO: 4. It is well known in the art that variants of a gene can alter both the structure and function of the protein encoded thereby in many different ways. As such the disclosed species are not representative of the structure and function of all members of the genus claimed

Therefore, the specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims.

***Enablement Rejections:***

The following is a quotation of the first paragraph of 35 U.S.C. 112:

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The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-10, 15-18, 23-24, 29 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a process of producing ergosta-5,7 dienol by culturing mutant *S. cerevisiae* Erg5 having deleted 22-desaturase gene and overexpressing said strain with HMG-CoA reductase gene comprising SEQ ID NO: 3 and squalene epoxidase gene of SEQ ID NO:7, does not reasonably provide enablement for process of producing ergosta-5,7 dienol by culturing any organisms having decreased  $\Delta 22$ -desaturase activity and increase of any HMG-CoA reductase activity which overexpress any squalene epoxidase or squalene epoxidase having 30% sequence identity to SEQ ID NO:8 and any HMG-CoA reductase. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, make and for use the invention commensurate in scope with these claims.

Claims 1-10, 15-18, 23-24 and 29 are so broad as to encompass any process of producing ergosta-5,7 dienol by culturing organisms having decreased  $\Delta 22$ -desaturase activity and which overexpress any squalene epoxidase or squalene epoxidase having 30% sequence identity to SEQ ID NO:8 and any HMG-CoA reductase. The scope of the claims is not commensurate with the enablement provided by the disclosure with regard to the extremely large number processes of producing ergosta-5,7 dienol by culturing organisms having decreased  $\Delta 22$ -desaturase activity, increased activity of any

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HMG-CoA reductase and which overexpress any squalene epoxidase or squalene epoxidase having 30% sequence identity to SEQ ID NO:8 broadly encompassed by the claims. Since the amino acid sequence of a protein determines its structural and functional properties, predictability of which changes can be tolerated in a protein's amino acid sequence and obtain the desired activity requires a knowledge of and guidance with regard to which amino acids in the protein's sequence, if any, are tolerant of modification and which are conserved (i.e. expectedly intolerant to modification), and detailed knowledge of the ways in which the proteins' structure relates to its function. However, in this case the disclosure is limited to a process of producing ergosta-5,7 dienol by culturing organisms having decreased  $\Delta 22$ -desaturase activity of a few desaturases and increase of a few HMG-CoA reductases and overexpression of squalene epoxidase (SEQ ID Nos:2, 4, 8, 10, 12, etc).

While recombinant and mutagenesis techniques are known, it is not routine in the art to screen for multiple substitutions or multiple modifications, as encompassed by the instant claims, and the positions within a protein's sequence where amino acid modifications can be made with a reasonable expectation of success in obtaining the desired activity/utility are limited in any protein and the result of such modifications is unpredictable (e.g., Whisstock, et al. Quarterly Rev. Biophy. 2003, 36, pp 307-340). In addition, one skilled in the art would expect any tolerance to modification for a given protein to diminish with each further and additional modification, e.g. multiple substitutions.

The specification does not support the broad scope of the claims which encompass any process of producing ergosta-5,7 dienol by culturing organisms having decreased  $\Delta 22$ -desaturase activity by inactivating any desaturase gene inactivated by any method and increase of any HMG-CoA reductase activity and overexpression of any squalene epoxidase or squalene epoxidase having 30% sequence identity to SEQ ID NO:8 because the specification does not establish: (A) regions of the protein structure which may be modified without effecting desaturase, HMG-CoA reductase and squalene epoxidase activity; (B) the general tolerance of desaturase, HMG-CoA reductase and squalene epoxidase to modification and extent of such tolerance; (C) a rational and predictable scheme for modifying any desaturase, HMG-CoA reductase squalene or epoxidase residues with an expectation of obtaining the desired biological function; and (D) the specification provides insufficient guidance as to which of the essentially infinite possible choices is likely to be successful.

Thus, applicants have not provided sufficient guidance to enable one of ordinary skill in the art to make and use the claimed invention in a manner reasonably correlated with the scope of the claims broadly including any process of producing ergosta-5,7 dienol by culturing organisms which  $\Delta 22$ -desaturase activity is decreased by any means and increase of any HMG-CoA reductase activity which overexpress any squalene epoxidase or squalene epoxidase having 30% sequence identity to SEQ ID NO:8 polypeptide with an enormous number of modifications of amino acid residues of a protein having amino acid sequence of SEQ ID NO: 8. The scope of the claims must bear a reasonable

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correlation with the scope of enablement (In re Fisher, 166 USPQ 19 24 (CCPA 1970)). Without sufficient guidance, determination of squalene epoxidase, having the desired biological characteristics is unpredictable and the experimentation left to those skilled in the art is unnecessarily, and improperly, extensive and undue. See In re Wands 858 F.2d 731, 8 USPQ2nd 1400 (Fed. Cir, 1988).

Applicants contend that claims 1-10, 15-18, 23-24 and 29 meet the enablement requirement of § 112, first paragraph because specification explains at pages 11-12 how ergosta-5,7 dienol is produced by using recombinant organism expressed with a full-length HMG-CoA-reductase gene encoding SEQ ID NO: 4 or squalene epoxidase of SEQ ID NO: 8. Applicant further argue that claimed methods is directed to the enzyme activity of the sterol biosynthetic enzymes not their percentage of identity. However, this is not persuasive as explained above the enzyme activity is correlated to the structure of the enzyme Furthermore; the scope of variant of genes encompassed in the instant claims are undefined. There are many way expression of squalene epoxidase and HMG-CoA reductase can be increased and  $\Delta 22$ -desaturase can be decreased in an organism. As the structure of the claimed squalene epoxidases and HMG-CoA reductases and desaturases that recite in the instant claims are not defined in any way, one of ordinary skill in the art would not be able to make and use any microorganism comprising said enzymes and require undue experimentation to first find what specific microorganisms comprising increased expression of squalene epoxidase and HMG-CoA reductase and decreased expression of desaturase

would be useful for the production of ergosta-5,7 dienol . Moreover, there are many ways in which a microorganisms' desaturase genes can be inactivated such as deletion, substitution of specific amino acid residues of DNA sequence or deletion of the whole genes, addition of inhibitors, modification of endogenous modulators, etc which would require substantial additional information that has not been provided by the specification. Also disclosed species of squalene epoxidase and HMG-CoA reductase (SEQ ID NO:8 and SEQ ID NO: ) are not representative of any variant of SEQ ID NO:8 and SEQ ID NO: 4. As such the disclosed species are not sufficient guidance to make and use any variant of SEQ ID NO:4 or 8. It is well known in the art that variation of a gene by mutation, substitution of nucleotide residues can alter both the structure and function of the protein encoded thereby in many different ways. As such the disclosed species are not sufficient to predict the effects of any mutation on the structure and function of all members of the genus claimed.

**THIS ACTION IS MADE FINAL.** See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the

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advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Mohammad Meah whose telephone number is 571-272-1261. The examiner can normally be reached on 8:30-5PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Nashaat T. Nashed can be reached on 571-272-0934. The fax phone number for the organization where this application or proceeding is assigned is 571-272-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see <http://pair-direct.uspto.gov>. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

Mohammad Younus Meah, PhD

Examiner, Art Unit 1652

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